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CARELLA, BYRNE, BAIN, GILFILLAN, CECCHI,
STEWART & OLSTEIN
5 BECKER FARM ROAD
ROSELAND, NJ 07068

EXAMINER	
STRZELECKA, TERESA E	
ART UNIT	PAPER NUMBER
1637	

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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	09/920,571	LASKEN ET AL.	
	Examiner	Art Unit	
	Teresa E Strzelecka	1637	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *01 June 2004 and 12 July 2004*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1,5-9,11-15,20-25,27,29,31-33,35-39,41,42,44-49,51-59 and 61* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1,5-9,11-15,20-25,27,29,31-33,35-39,41,42,44-49,51-59 and 61* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

Continued Examination Under 37 CFR 1.114

1. A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicant's submission filed on July 12, 2004 has been entered. Applicants' amendment filed June 1, 2004 has been entered.

2. Claims 1, 5-9, 11-15, 20-25, 27, 29-59 and 61 were previously pending. Applicants cancelled claims 30, 34, 40, 43, and 50 and amended claims 1, 29 and 56. Claims 1, 5-9, 11-15, 20-25, 27, 29, 31-33, 35-39, 41, 42, 44-49, 51-59 and 61 are pending and will be examined. Applicants' amendments and claim cancellations overcame the following rejections: rejection of claims 1, 5-7, 11-15, 20-25, 27, 29, 31, 33, 35-40, 42, 44, 45, 48-54, 57, 58 and 61 under 35 U.S.C. 103(a) over Lizardi-1 (U.S. Patent No. 5,854,033) and Lizardi-2 (U.S. patent No. 6,124,120); rejection of claims 8 and 9 under 35 U.S.C. 103(a) over Lizardi-1 (U.S. Patent No. 5,854,033) and Lizardi-2 (U.S. patent No. 6,124,120) in view of Sorge et al. (U.S. Patent No. 5,599,921); rejection of claims 32, 41, 46, 47 and 59 under 35 U.S.C. 103(a) over Lizardi-1 (U.S. Patent No. 5,854,033) and Lizardi-2 (U.S. patent No. 6,124,120) in view of Skerra et al.; rejection of claims 30, 34 and 43 under 35 U.S.C. 103(a) over Lizardi-1 (U.S. Patent No. 5,854,033) and Lizardi-2 (U.S. patent No. 6,124,120) in view of Cummins et al.; rejection of claims 55 and 56 under 35 U.S.C. 103(a) over Lizardi-1 (U.S. Patent No. 5,854,033) and Lizardi-2 (U.S. patent No. 6,124,120) in view of Sorge et al. (U.S. Patent No. 5,556,772).

3. Applicants' arguments regarding the art rejections are moot in view of new grounds of rejection.

Priority

4. Currently pending claims have the priority date which is the filing date of the instant application, July 31, 2001. The priority application, 09/605,592 does not provide support for the limitation of at least one nucleotide (dNTP) rendering TS-DNA resistant to nuclease activity following incorporation into the TS-DNA (see also written description rejection below).

Claim Objections

5. Applicant is advised that should claim 53 be found allowable, claim 54 will be objected to under 37 CFR 1.75 as being a substantial duplicate thereof. When two claims in an application are duplicates or else are so close in content that they both cover the same thing, despite a slight difference in wording, it is proper after allowing one claim to object to the other as being a substantial duplicate of the allowed claim. See MPEP § 706.03(k).

Both claims are drawn to DNA polymerase being bacteriophage ϕ29 DNA polymerase and multiple primers resistant to exonuclease activity.

6. Claim 61 is objected to under 37 CFR 1.75(c), as being of improper dependent form for failing to further limit the subject matter of a previous claim. Applicant is required to cancel the claim(s), or amend the claim(s) to place the claim(s) in proper dependent form, or rewrite the claim(s) in independent form.

Claim 61, dependent from claim 56, is drawn to the DNA polymerase being ϕ29 DNA polymerase. Claim 56 is drawn to a Markush group of Taq, Tfl, Tth and eucaryotic DNA polymerase alpha polymerases, but does not contain the ϕ29 DNA polymerase.

Claim Rejections - 35 USC § 112

7. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

8. Claims 1, 5-9, 11-15, 20-25, 27, 29, 31-33, 35-39, 41, 42, 44-49, 51-59 and 61 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

MPEP 2163.06 (I) states:

> If new matter is added to the claims, the examiner should reject the claims under 35 U.S.C. 112, first paragraph - written description requirement. *In reRasmussen*, 650 F.2d 1212, 211 USPQ 323 (CCPA 1981).

The claims as presented in an amendment filed June 12, 2002 introduced new matter, namely, a limitation of at least one nucleotide (dNTP) rendering TS-DNA resistant to nuclease activity following incorporation into the TS-DNA. There is no support in the priority application, 09/605,592, in the current specification or in the originally filed claims for incorporation of dNTPs into an amplification product, the dNTPs conferring nuclease resistance to the amplification product. On page 6 of the specification, last paragraph, continued on page 7, Applicants describe primers which are resistant to enzyme degradation. Figure 5 shows results of amplification with nuclease resistant primers (legend on page 8, lines 18). On page 17, lines 3-6, on page 21, lines 20-27, on page 22, lines 19-28 and in Example 5 on page 37 Applicants describe using exonuclease resistant primers. On page 25, lines 7-18, Applicants describe solid phase synthesis of exonuclease-resistant primers containing phosphorothioate residues.

Therefore, the disclosure as originally filed does not describe incorporation of nuclease-resistant dNTPs into amplified nucleic acid during enzymatic synthesis of the nucleic acid, and for this reason the claims do not comply with the written description requirement.

9. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

10. Claim 35 rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 35 recites the limitation "said at least one nucleotide" in line 1/2. There is insufficient antecedent basis for this limitation in the claim. Claim 1, from which claim 35 depends, does not contain a limitation of "at least one nucleotide".

Claim Interpretation

11. Applicants defined the term "random primers" on page 13, lines 20-26: "... As used herein, the term "random" means that said oligonucleotide primers (P1) have nucleotide sequences unrelated to the nucleotide sequences of the amplification target circle (ATC) that acts as template for amplification. The result of such a random relationship is that the locations on the ATC at which said random primers hybridize will also be random."

Claim Rejections - 35 USC § 103

12. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

13. Claims 1, 5-9, 11, 13, 15, 20-25, 27, 29, 31, 33, 35, 38, 39, 41, 44-49, 51-56 and 61 are rejected under 35 U.S.C. 103(a) as being unpatentable over Lizardi (U.S. Patent No. 5,854,033; cited in the previous office action), Landers et al. (U.S. Patent No. 6,703,228) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989).

A) Regarding claim 1, Lizardi teaches a method of amplification comprising contacting multiple single stranded non-circular random oligonucleotide primers (P1), one or more amplifIcation target circles (ATCs), a DNA polymerase and multiple deoxynucleoside triphosphates (dNTP), under conditions promoting said contacting, wherein an ATC hybridizes to a plurality of said P1 primers, wherein said conditions promote replication of the amplification target circle by extension of the P1 primers to form multiple tandem sequence DNA (TS-DNA) products and wherein at least one such dNTP renders the TS-DNA resistant to nuclease activity following incorporation thereinto (Lizardi teaches amplification of circular DNA molecule by a rolling circle method. The rolling circle amplification (RCA) involves hybridization (= contacting) of a primer (P1) to amplification target circles (ATC) followed by amplification using strand-displacing DNA polymerase, resulting in a DNA molecule with multiple repeats of the ATC, usually referred to as tandem sequences DNA (TS-DNA) (column 19, lines 20-31). In one embodiment of the amplification, strand displacement cascade amplification, (SDCA), secondary and tertiary primers are used, with sequences complementary to the ATC (col. 25, lines 36-49). The SDCA can be performed simultaneously with RCA, resulting in exponential amplification (col. 28, lines 8-18; col. 26, lines 61-66). Therefore, Lizardi teaches the limitation of multiple P1 primers. Lizardi teaches dNTPs (col. 36, lines 50, 51).

Regarding claims 5-7, Lizardi teaches primers from 10 to 35 nucleotides long (col. 10, line 14), therefore anticipating the limitations of the primers being 2 to 50, 2 to 35 or 2 to 10 nucleotides in length.

Regarding claim 11, Lizardi teaches ATC being a circular, single-stranded DNA molecule, (col. 9, lines 25-29).

Regarding claim 20, Lizardi teaches ATC being a circular, single-stranded DNA molecule, containing between 40 to 1,000 nucleotides (col. 9, lines 25-29), anticipating the limitation of ATC being no larger than 10,000 nucleotides in size.

Regarding claims 22 and 23, Lizardi teaches ATC being a circular, single-stranded DNA molecule, containing between 40 to 1,000 nucleotides (col. 9, lines 25-29), anticipating the limitations of ATC being no larger than about 1,000 nucleotides and no larger than about 100 nucleotides in size.

Regarding claim 24, Lizardi teaches that ATC can be derived from a single-stranded bacteriophage (col. 35, lines 50-59).

Regarding claim 27, Lizardi teaches that radioactive nucleotides can be used in the amplification (col. 21, lines 22-25).

Regarding claim 31, Lizardi teaches that primers may include modified nucleotides to make them exonuclease-resistant (col. 10, lines 24-28; col. 13, lines 27-31). Therefore Lizardi teaches exonuclease activity.

Regarding claim 33, Lizardi teaches adding exonuclease to digest unligated circles (col. 10, lines 28-33; col. 24, lines 41-61).

Regarding claim 35, Lizardi teaches that modified nucleotides can be used in the amplification (col. 21, lines 22-25).

Regarding claims 38 and 39, Lizardi teaches primers which include modified nucleotides to make them exonuclease-resistant (col. 10, lines 24-28; col. 13, lines 27-31).

Regarding claims 44, 45 and 47, Lizardi teaches that phophorothioate nucleotides are positioned at the 5'-end of the primer to make it exonuclease-resistant (col. 10, lines 24-28; col. 13, lines 27-31). Therefore Lizardi anticipates the limitations of an exonuclease-resistant primer containing at least one nucleotide which makes it resistant to exonuclease activity, a modified nucleotide and a phosphorothioate nucleotide.

Regarding claim 48, Lizardi teaches three or four phosphorothioate nucleotides (col. 10, lines 24-28; col. 13, lines 27-31).

Regarding claim 49, Lizardi teaches the phosphorothioate nucleotides being at the 5' end of the primer (col. 10, lines 24-28; col. 13, lines 27-31).

Regarding claims 51, 52 and 61, Lizardi teaches the following DNA polymerases to be used: bacteriophage ϕ 29 DNA polymerase, phage M2 DNA polymerase, VENT DNA polymerase, Klenow fragment of DNA polymerase I, T5 DNA polymerase, PRD1 DNA polymerase, T4 DNA polymerase holoenzyme (col. 17, lines 66-67, col. 18, lines 1-11). Therefore, since the claim language links 3',5'-exonuclease activity with these enzymes, and Lizardi specifically teaches them, Lizardi inherently teaches polymerases with 3' -> 5' exonuclase activity.

Regarding claims 53 and 54, Lizardi teaches bacteriophage ϕ 29 DNA polymerase (col. 17, lines 66-67, col. 18, lines 1-11) and exonuclease-resistant primers (col. 10, lines 24-28; col. 13, lines 27-31).

Regarding claims 36 and 37, Lizardi teaches oligonucleotides attached to solid support, including glass (col. 14, lines 34-43, 65-67; col. 15, lines 1-10).

B) Lizardi does not teach random primers, nucleotides which confer nuclease resistance to an amplification product, duplex DNA, denaturation of duplex DNA, DNA larger than 10,000 nucleotides, DNA with unknown sequence, dNTPs being phosphorothioate nucleotides, modified nucleotide being a 3'-terminal nucleotide or a DNA polymerase which does not exhibit 3' -> 5' exonuclease activity.

C) Regarding claim 1, Landers et al. teach generation of reduced complexity genomes by amplification of genomic double-stranded DNA circles (YACs) with multiple arbitrary (= random) primers (col. 17, lines 28-42 and 60-64).

Regarding claims 8 and 9, Landers et al. teach that the sequence of the random primers contains the N_x residues of the DOP-PCR primers (col. 17, lines 35-39). Landers et al. teach DOP-PCR primers containing x N residues, where x is an integer from 0 to 9, therefore Landers et al. teach hexamers and octamers.

Regarding claim 13, Landers et al. teach YACs (Yeast Artificial Chromosomes), which are double-stranded circles (col. 17, lines 64, 65).

Regarding claim 15, Landers et al. teach denaturation of double-stranded DNA during PCR amplification (col. 14, lines 14-36; col. 62, lines 36-43; col. 63, lines 16-25).

Regarding claim 21, Landers et al. teach YACs (Yeast Artificial Chromosomes) carrying inserts of 300,000-400,000 base pairs (col. 17, lines 64-66), therefore Landers et al. teach ATCs larger than 10,000 nucleotides in size.

Regarding claim 25, Landers et al. teach amplification of unknown sequences (col. 17, lines 31-34).

Regarding claims 55 and 56, Landers et al. teach amplification using Taq DNA polymerase (col. 14, lines 51-54). Therefore, since the claim language connects 3',5'-exonuclease activity with

Taq DNA polymerase, and Landers et al. teach Taq DNA polymerase, they inherently teach a DNA polymerase without 3'5'-exonuclease activity.

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have included random primers and DNA molecules of Landers et al. in the method of Lizardi. The motivation to do so, provided by Landers et al., would have been that random primers allowed for amplification of unknown DNA sequences (col. 17, lines 31-34), and using double-stranded DNA allowed for reducing complexity of genomic samples for purposes of genotyping related to screening of populations for diseases (col. 10, lines 13-24).

D) Landers et al. do not teach nucleotides which confer nuclease resistance to an amplification product, dNTPs being phosphorothioate nucleotides, modified nucleotide being a 3'-terminal nucleotide.

E) Regarding claims 1 and 29, Eckstein et al. teach that deoxynucleoside 5'-O-(1-thiotriphosphates), or phoshorothioates, are substrates for DNA and RNA polymerases (Abstract; page 97, first paragraph).

Regarding claim 41, Eckstein et al. teach exonuclease III with 3',5'-exonuclease activity (page 97, fourth paragraph).

Regarding claims 46 and 47, Eckstein et al. teach that incorporation of single phosphorothioate group at the 3' end of a DNA strand prevents its degradation by exonuclease III, an enzyme with 3',5' activity (page 97, fourth paragraph).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used phosphorothioate dNTPs of Eckstein et al. in the amplification method of Lizardi and Landers et al. The motivation to do so, provided by Eckstein et al., would have been

that phosphorothioate containing DNA was resistant to degradation by nucleases and the sulfur atom conferred many favorable chemical properties (Abstract).

14. Claims 12, 36 and 37 are rejected under 35 U.S.C. 103(a) as being unpatentable over Lizardi (U.S. Patent No. 5,854,033; cited in the previous office action), Landers et al. (U.S. Patent No. 6,703,228) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989) as applied to claim 1 above, and further in view of Rothberg et al. (U.S. Patent No. 6,274,320).

A) Claim 12 is drawn to a duplex DNA circle having at least one nick, claim 36 is drawn to at least one P1 primer attached to a solid support and claim 37 is drawn to the solid support being glass or plastic.

B) Lizardi teaches oligonucleotides attached to solid support, the support being glass or plastic. Lizardi, Landers et al. and Eckstein et al. do not teach duplex DNA circles with at least one nick or primers attached to solid support.

C) Regarding claim 12, Rothberg et al. teach templates are open or closed circles (col. 3, lines 57-60), and nicked double-stranded circles (col. 12, lines 26-35).

Regarding claim 36, Rothberg et al. teach amplification of circular templates by rolling circle amplification using primers attached to a solid support (Fig. 1; col. 3, lines 66,67; col. 4, lines 1-10 and 29-37; col. 5, lines 6-20; col. 11, lines 54-67; col. 12, lines 1-9).

Regarding claim 37, Rothberg et al. teach solid supports being a DNA chip or a glass slide (col. 19, lines 40-43) or an optical fiber (col. 20, lines 15-18).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used primers attached to a solid support of Rothberg et al. in the method of Lizardi, Landers et al. and Eckstein et al. The motivation to do so, provided by Rothberg et al., would have been that amplification of circular templates on solid support allowed for determination

of nucleic acid sequence without the need for cloning the templates and determination of rare nucleic acids with high sensitivity (col. 5, lines 22-28).

15. Claims 14, 57 and 58 are rejected under 35 U.S.C. 103(a) as being unpatentable over Lizardi (U.S. Patent No. 5,854,033; cited in the previous office action), Landers et al. (U.S. Patent No. 6,703,228) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989) as applied to claim 1 above, and further in view of Navarro et al. (J. Virol. Meth., vol. 56, pp. 59-66, 1996).

A) Claim 14 is drawn to ATC being single stranded RNA circle, claim 57 is drawn to DNA polymerase being a reverse transcriptase and claim 58 is drawn to the ATC being RNA and DNA polymerase being a reverse transcriptase.

B) Lizardi, Landers et al. and Eckstein et al. do not teach RNA circles or a reverse transcriptase.

C) Navarro et al. teach amplification of circular RNA viroids using random hexamers and AMV reverse transcriptase (Fig. 1; page 59, first paragraph; page 60, paragraphs 4 and 5; page 61, first paragraph).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used the amplification of RNA circles of Navarro et al. in the method of Lizardi, Landers et al. and Eckstein et al. The motivation to do so, provided by Navarro et al., would have been that amplification of circular pathogenic RNA provided means of cloning the RNAs from small amounts of sample with unknown sequence (page 60, second paragraph).

16. Claims 32, 42 and 59 are rejected under 35 U.S.C. 103(a) as being unpatentable over Lizardi (U.S. Patent No. 5,854,033; cited in the previous office action), Landers et al. (U.S. Patent No. 6,703,228) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989) as applied to

claims 1, 31 and 38 above, and further in view of Skerra (Nucleic Acids Research, Vol. 20, pp. 3551-3554, 1992; cited in the previous office action).

A) Claims 32 and 42 are drawn to a polymerase with 3'-> 5' exonuclease activity and claim 59 to the use of a mixture of primers sensitive to and resistant to exonuclease activity.

B) Lizardi, Landers et al. and Eckstein et al. do not teach primers resistant to 3'-> 5' exonuclease activity, the resistance being conferred by a phosphorothioate nucleotide at the 3'-end of the primer or the use of a mixture of exonuclease-sensitive and exonuclease-resistant primers in the amplification reaction.

C) Skerra teaches that incorporation of a phosphorothioate nucleotide at the 3'-end of the primer renders it inactive to the 3'-> 5' exonuclease activity of DNA polymerases such as Vent and Pfu. The reference also teaches use of a mixture of exonuclease-sensitive and exonuclease-resistant primers in the amplification reaction (Abstract; page 3553; Fig. 2).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used primers of Skerra with phosphorothioate nucleotides at the 3'-end in the amplification method of Lizardi, Landers et al. and Eckstein et al. The motivation to do so, provided by Skerra, would have been that the 3'-end phosphorothioate nucleotide rendered the primers resistant to 3'-> 5' exonuclease activity of the polymerase used in the reaction, resulting in an improved yield of the amplification product (page 3553, third paragraph).

Rejections based on the Dean et al. reference

17. Claims 1, 5-8, 11, 13, 15, 20, 21, 24, 27, 29, 31-33, 35, 38, 39, 41, 42, 44-49, 51-54 and 61 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dean et al. (Genome Res., vol. 11, pp. 1095-1099, June 1, 2001; cited in the IDS) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989).

A) Regarding claim 1, Dean et al. teach amplification of vector DNAs, the method comprising contacting M13 circular DNA (ATC) with random hexamer primers, dNTPs and DNA polymerase under conditions promoting hybridization and replication of the ATC to form multiple tandem sequence DNA (Fig. 1; page 1095, fourth paragraph; page 1099, paragraphs 3 and 4).

Regarding claims 5-8, Dean et al. teach hexamer primers, therefore they teach primers which are within the range of 2 to 50, 2 to 35 or 2 to 10 nucleotides in length.

Regarding claim 11, Dean et al. teach single-stranded M13 circle (page 1099, third paragraph).

Regarding claim 13, Dean et al. teach double-stranded plasmids pUC19 and pUC18 library (page 1099, fifth and sixth paragraphs).

Regarding claim 15, Dean et al. teach denaturation of double-stranded DNA (page 1099, fifth and sixth paragraphs).

Regarding claim 20, Dean et al. teach M13 and pUC19 being no larger than 10,000 base pairs in size (Fig. 3).

Regarding claim 21, Dean et al. teach amplification of BACs and cosmids, therefore they teach amplification of circles larger than 10,000 nucleotides in size (page 1098, first paragraph).

Regarding claim 24, Dean et al. teach single-stranded bacteriophage M13 circle (page 1099, third paragraph).

Regarding claims 27 and 35, Dean et al. teach radiolabeled, i.e., modified, dNTPs (page 1099, fourth paragraph).

Regarding claims 31-33, 41 and 42, Dean et al. teach 3',5' exonuclease activity of the ϕ29 DNA polymerase, which is added to the reaction mixtures (page 1097, first paragraph; page 1099, fifth and sixth paragraphs).

Regarding claims 38 and 39, Dean et al. teach primers resistant to exonuclease activity (page 1097, first paragraph; page 1099, fifth and sixth paragraphs).

Regarding claims 44 and 45, Dean et al. teach primers resistant to exonuclease activity containing modified nucleotides (page 1097, first paragraph).

Regarding claim 46, Dean et al. teach primers resistant to exonuclease activity containing modified nucleotides at the 3' terminus (page 1097, first paragraph).

Regarding claim 47, Dean et al. teach primers resistant to exonuclease activity containing phosphorotioate nucleotides (page 1097, first paragraph).

Regarding claim 48, Dean et al. teach primers resistant to exonuclease activity containing two modified nucleotides (page 1097, first paragraph).

Regarding claim 49, Dean et al. teach primers resistant to exonuclease activity containing a modified nucleotide at a second base from the 3' end (page 1097, first paragraph).

Regarding claims 51-54 and 61, Dean et al. teach 3',5' exonuclease activity of the ϕ29 DNA polymerase, which is added to the reaction mixtures (page 1097, first paragraph; page 1099, fifth and sixth paragraphs).

B) Dean et al. do not teach amplification using dNTPs which render the amplification product resistant to nuclease activity.

C) Regarding claims 1 and 29, Eckstein et al. teach that deoxynucleoside 5'-O-(1-thiotriphosphates), or phoshorothioates, are substrates for DNA and RNA polymerases (Abstract; page 97, first paragraph).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used phosphorothioate dNTPs of Eckstein et al. in the amplification method of Dean et al. The motivation to do so, provided by Eckstein et al., would have been that

phosphorothioate containing DNA was resistant to degradation by nucleases and the sulfur atom conferred many favorable chemical properties (Abstract).

18. Claims 12, 22, 23, 36 and 37 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dean et al. (Genome Res., vol. 11, pp. 1095-1099, June 1, 2001; cited in the IDS) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989) as applied to claim 1 above, and further in view of Rothberg et al. (U.S. Patent No. 6,274,320).

A) Claim 12 is drawn to a duplex DNA circle having at least one nick, claim 22 is drawn to an ATC being no larger than 1,000 nucleotides, claim 23 is drawn to ATC being no larger than 100 nucleotides, claim 36 is drawn to at least one P1 primer attached to a solid support and claim 37 is drawn to the solid support being glass or plastic.

B) Neither Dean et al. nor Eckstein et al. teach duplex DNA circles with at least one nick, circles smaller than 1000 or 100 nucleotides or primers attached to solid support.

C) Regarding claim 12, Rothberg et al. teach templates are open or closed circles (col. 3, lines 57-60), and nicked double-stranded circles (col. 12, lines 26-35).

Regarding claims 22 and 23, Rothberg et al. teach circular templates 10-1000, 10-200, 10-100, 10-50 or 20-40 nucleotides in length (col. 3, lines 57-63), anticipating the limitations of templates smaller than 1000 or 100 nucleotides in size.

Regarding claim 36, Rothberg et al. teach amplification of circular templates by rolling circle amplification using primers attached to a solid support (Fig. 1; col. 3, lines 66,67; col. 4, lines 1-10 and 29-37; col. 5, lines 6-20; col. 11, lines 54-67; col. 12, lines 1-9).

Regarding claim 37, Rothberg et al. teach solid supports being a DNA chip or a glass slide (col. 19, lines 40-43) or an optical fiber (col. 20, lines 15-18).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used primers attached to a solid support of Rothberg et al. in the method of Dean et al. and Eckstein et al. The motivation to do so, provided by Rothberg et al., would have been that amplification of circular templates on solid support allowed for determination of nucleic acid sequence without the need for cloning the templates and determination of rare nucleic acids with high sensitivity (col. 5, lines 22-28).

19. Claims 14, 25, 57 and 58 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dean et al. (Genome Res., vol. 11, pp. 1095-1099, June 1, 2001; cited in the IDS) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989) as applied to claim 1 above, and further in view of Navarro et al. (J. Virol. Meth., vol. 56, pp. 59-66, 1996).

A) Claim 14 is drawn to ATC being single stranded RNA circle, claim 25 is drawn to an ATC with unknown sequence, claim 57 is drawn to DNA polymerase being a reverse transcriptase and claim 58 is drawn to the ATC being RNA and DNA polymerase being a reverse transcriptase.

B) Neither Dean et al. nor Eckstein et al. teach RNA circles or a reverse transcriptase.

C) Regarding claims 14, 57 and 58, Navarro et al. teach amplification of circular RNA viroids using random hexamers and AMV reverse transcriptase (Fig. 1; page 59, first paragraph; page 60, paragraphs 4 and 5; page 61, first paragraph).

Regarding claim 25, Navarro et al. teach amplification of templates of unknown sequence (page 60, second paragraph; page 64, third paragraph).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used the amplification of RNA circles of Navarro et al. in the method of Lizardi, Landers et al. and Eckstein et al. The motivation to do so, provided by Navarro et al., would have

been that amplification of circular pathogenic RNA provided means of cloning the RNAs from small amounts of sample with unknown sequence (page 60, second paragraph).

20. Claims 55 and 56 are rejected under 35 U.S.C. 103(a) as being unpatentable over Dean et al. (Genome Res., vol. 11, pp. 1095-1099, June 1, 2001; cited in the IDS) and Eckstein et al. (Trends in Bioch. Sci., vol. 14(3), pp. 97-100, 1989) as applied to claim 1 above, and further in view of Sorge et al. (U.S. Patent No. 5,556,722; cited in the previous office action).

A) Claim 55 is drawn to DNA polymerase without the 3'->5' exonuclease activity, and claim 56 to specific DNA polymerases not exhibiting this activity (e.g. Taq, Tfl, etc.)

B) Neither Dean et al. nor Eckstein et al. teach DNA polymerases without the 3'->5' exonuclease activity (exo(-)) or Taq polymerase.

C) Sorge et al. teaches Taq DNA polymerase which lacks 3'->5' exonuclease activity (col. 5, lines 30-61).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time of the invention to have used exo(-) Taq DNA polymerase in the method of Dean et al. and Eckstein et al. The motivation to do so, provided by Sorge et al., would have been that Taq polymerase was highly processive, with an extension rate of more than 60 nucleotides per second (col. 5, lines 62-64).

21. No claims are allowed.

Conclusion

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Teresa E Strzelecka whose telephone number is (571) 272-0789. The examiner can normally be reached on M-F (8:30-5:30).

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on (571) 272-0782. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

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August 19, 2004

Teresa Strzelecka
Teresa Strzelecka
Patent Examiner